Applied Medical Research, 2019
VOL 6, NO. 1, PAGE 1 – 7
DOI: doi.org/10.47363/AMR/2019(6)165

ORIGINAL RESEARCH

Open Access

Influence of vitamin D status on atherogenic profile of apparently healthy young adults

Oyebola Oluwagbemiga Sonuga[1], Ololade Oluwatobi Eluyera[1], Ayobola Abimbola Sonuga[2]
[1]Department of Chemical Pathology, Babcock University, Ogun, Nigeria
[2]Department of Science Laboratory Technology (Biochemistry Option), Ekiti State University, Nigeria

ABSTRACT

Background: Cardiovascular disease (CVD) is the most common group of non-communicable diseases and responsible for major morbidity and mortality worldwide. Low serum concentrations of 25(OH) vitamin D (vitamin D status) are associated with CVD risk factors and might predict the occurrence of cardiovascular events through its influence on the progression of atherogenesis**.**
Aim: This study aimed to determine the association between vitamin D status and some markers of atherogenesis in apparently healthy youths.
Methods: 150 young adults (students) aged 18–25 years were recruited from tertiary educational institutions in Ibadan, Nigeria, in 2018, and grouped into two based on their vitamin D status; Group A [25(OH) vitamin D ≥30 ng/ml, i.e., sufficient] and Group B [25(OH) vitamin D < 30 ng/ml, i.e., insufficient/deficient]. Anthropometric data, plasma glucose, total cholesterol (TC), high-density lipoprotein-cholesterol (HDL-C), triglycerides, low-density lipoprotein-cholesterol (LDL-C); and serum insulin, highly sensitive C-reactive protein (hsCRP), apolipoprotein B (Apo B), apolipoprotein A1 (ApoA1), ApoB/ApoA1 ratio, and lipoprotein (a) [Lp (a)] were determined by standard methods and compared between the two groups.
Results: Statistical analysis was performed using SPSS version 20.0. Student's *t*-test and Pearson correlation were used to compare and assess the relationship between normally distributed variables, while the Mann–Whitney U test and Spearman rank correlation were used for non-Gaussian variables. Results showed that there was no significant statistical difference in the compared variables between the two groups, except for HDL-C which was significantly lower in the vitamin D deficient group compared to vitamin D sufficient group. The vitamin D sufficient participants were also taller than the deficient/insufficient group. There was a weak positive correlation between vitamin D and HDL-C; and a weak negative correlation between vitamin D and hs-CRP in the vitamin D sufficient group.
Conclusion: This present study supports evidence from previously published works that reduced vitamin D status could increase both inflammation and dyslipidemia, which are both well-known risk factors for incident cardiovascular disease. It also supports existing evidence suggesting that adequate vitamin D status could reduce the risks of overt adult cardiovascular disease.

ARTICLE HISTORY

Received April 09, 2019
Accepted July 19, 2019
Published August 05, 2019

KEYWORDS

Vitamin D; atherogenic markers; cardiovascular diseases; healthy young adults

Introduction

Cardiovascular disease (CVD) is the leading cause of death worldwide and hypertension is the leading associated risk factor [1]. It has been suggested that the prevalence of CVD, especially hypertension, is increasing rapidly in sub-Saharan African countries, such as Nigeria [2], where the current prevalence is already as high as that seen in developed countries [3]. The number of adults living with hypertension by 2025 is predicted to increase by about 60% to a

Contact Oyebola Oluwagbemiga Sonuga ✉ oyebolasonuga@yahoo.com Department of Chemical Pathology, Babcock University, Ogun, Nigeria.

total of 1.56 billion and more than 80% of the global burden of CVD will occur in low- and middle-income countries, including sub-Saharan African countries like Nigeria [4].

Early intervention aimed at modifiable risk factors can help prevent or delay CVDs and other co-morbid diseases, such as type 2 diabetes [5]. The underlying changes that lead to atherogenesis begin years before the overt disease develops and these changes include small changes in biomarkers of inflammation, glycemic index, lipid metabolism, and blood pressure [6,7]. In general, higher levels of atherogenic biomarkers are considered to be independent risk factors for CVD [8]. Young adults are a key target subgroup for substantial preventive healthcare strategies. However, atherogenic biomarkers are not often monitored in healthy young adults, remaining undiagnosed until the effects of the advanced pathologic condition become apparent in later life.

Dyslipidemia, defined as raised levels of TC, triglycerides (TG), and low-density lipoprotein-cholesterol (LDL-C) and decreased levels of high-density lipoprotein-cholesterol (HDL-C), has been identified to be important risk factors of atherosclerosis and CVD [9]. It has been confirmed that lowering serum cholesterol results in a reduction in cardiovascular morbidity. Lipoprotein (a) is a LDL—like substance with a single molecule of apolipoprotein B100 (apoB100) covalently linked by disulfide bridges, in a 1:1 molar ratio, to apolipoprotein(a) [apo(a)]. It is pro-atherogenic and its pro-thrombotic effects are due to its similar structure both to LDL-C and to plasminogen [10], explaining its association with the pathogenesis of CVD. Elevated Lp(a) levels were proven as a marker of increased CVD risk in numerous epidemiological and genetic studies [11,12]. Apolipoprotein B is the structural protein found in all the atherogenic lipoproteins (VLDL-C, IDL-C, LDL-C, and Lp(a)) and modulates the transportation of lipids from the liver and gut to peripheral tissues. Apo B is not simply a measurement of risk, such as LDL-C or non-HDL-C, but it is itself causative for the progression of atherosclerosis. The reverse cholesterol transport (a multi-step process resulting in the net movement of cholesterol from peripheral tissues back to the liver, where it is converted to cholesteryl esters by lecithin cholesterol acyltransferase) and formation of HDL-C are the basic role of Apolipoprotein-A1. Low levels of Apo A1 have been identified as a risk factor in the development and progression of coronary damage [13]. The higher the value of the apoB/apoA-1 ratio, the higher the value of circulating plasma cholesterol which is then more likely to be deposited in the arterial wall, provoking atherogenesis and increased risk of subsequent CVD events [14]. The lower the apoB/apoA-I ratio, the lower is the challenge of cholesterol to the periphery, the greater is the reverse cholesterol transport and other beneficial functions, and the lower the risk of CVD events [14].

C-reactive protein (CRP), a member of the pentraxin family, is an acute phase reactant synthesized mainly by the liver (and in many other tissues). Serum CRP concentrations are elevated in response to acute infections, inflammatory conditions, and trauma. It also appears to have predictive value in both stable and unstable angina as well as in the chronic phase after myocardial infarction [15]. In addition, increased highly sensitive CRP (hsCRP) concentrations are associated with multiple risk factors for CVD, including obesity, insulin resistance, diabetes mellitus, and hypertension; and it has demonstrated significant predictive value for risk of metabolic syndrome [16,17]. The American Heart Association/Centre for Disease Control Working Group on markers of inflammation in CVD has classified the risk of developing CVD based on serum levels of hsCRP as; low risk (<1 mg/l), moderate risk (1–3 mg/l), and high risk (>3 mg/l).

Vitamin D is a fat-soluble vitamin that functions as a steroid hormone precursor and can be obtained from the diet (vitamin D2 in fortified foods, mushrooms; vitamin D3 in fish oil, liver, butter, egg yolk, etc); or from endogenous synthesis in the skin with exposure to ultraviolet B light (vitamin D3-cholecalciferol). Traditionally, vitamin D is known for its role in calcium and phosphorus homeostasis. However, recent research has shown that vitamin D is a prehormone of calcitriol which has numerous extra skeletal roles. Many cell types, including vascular smooth muscle cells, endothelial cells, and cardiomyocytes, produce 1α-hydroxylase, which converts 25-hydroxyvitamin D to calcitriol, the main ligand of the vitamin D receptor. Calcitriol has been shown to inhibit vascular smooth muscle cell proliferation, down-regulate the renin-angiotensin-aldosterone system (RAAS), decrease coagulation, and to exhibit anti-inflammatory properties. Epidemiological studies provide evidence that low serum concentrations of 25(OH) vitamin D are associated with CVD risk and can predict the occurrence of cardiovascular events, including myocardial infarction and stroke

[18,19]. In fact, vitamin D deficiency may lead to elevated levels of atherogenic markers [20]. A study by Reis et al. [21] showed that low vitamin D status in adolescents was strongly associated with increased risks for fasting hyperglycemia, hypertension, and metabolic syndrome. In 1,739 Framingham Offspring Study participants, the multivariable-adjusted risk for incident CVDs such as myocardial infarction, coronary insufficiency, and heart failure was higher in individuals with insufficient or deficient 25(OH) D concentrations compared to those with sufficient 25(OH) D concentrations [22]. Giovannucci reported that, after adjustment for cardiac risk factors, individuals with low 25(OH) D concentrations (<10 ng/ml) were about three times more likely to die of heart failure and five times more likely to die of sudden cardiac death, compared with individuals with 25(OH) D concentrations of at least 30 ng/ml [23]. There are also growing bodies of data from studies of young adults and elderly persons that vitamin D deficiency is an unrecognized and prevalent health problem [24,25].

However, whether vitamin D status influences atherogenic risk markers, thereby increasing CVD risk has not been thoroughly studied in Nigeria, especially in young adults. This study aimed to determine relationships if any between vitamin D status and recognized risk markers of atherogenesis in apparently healthy young adults.

Materials and Methods

Study design

This is a cross-sectional study carried out in 2018, among age-matched young adults (students) from tertiary educational institutions in Ibadan over a period of 8 months. A total of 150 apparently healthy participants with an age range from 18 to 25 years were enrolled and grouped into two based on their vitamin D status; Group A (young adults with serum 25(OH) D concentrations ≥30 ng/ml) and Group B (young adults with serum 25(OH) D concentrations <30 ng/ml). It is important to note that all the participants of this study were from South-Western Nigeria and exposed to the same pattern of diet and though with a different religious background, wore the same pattern of clothing. Vitamin D status [assessed by serum 25(OH) D concentrations], was defined as Deficient (<10 ng/ml); Insufficient (10n g/ml–30 ng/ml); and Sufficient (30 ng/ml–100 ng/ml) according to manufacturer's manual (CALBIOTECH Diagnostics) [26]. Ethical approval was obtained from the University of Ibadan/ University College Hospital, Ibadan Health Research Ethics Committee. Privacy and confidentiality of the participants were guaranteed by the coding of the data to ensure the anonymity of the participants.

Data collection

A data collection form (questionnaire) which contained items on the demographic characteristics, clinical measurements, medical history, diet history, and results of the laboratory analysis of participants was used for study recruitment. Data collection was by personal interview and an informed written consent form was obtained from all participants after educating them on the purpose and relevance of the study. The clinical measurements that included weight (Kg) and height (m) were obtained with the participants wearing light clothing without shoes, by stadiometer, and the stadiometer's headpiece, respectively. BMI was calculated as weight/height^2 (kg/m^2). Waist circumference was measured in "cm" at the midpoint between the lower margin of the least palpable rib and the top of the iliac crest (umbilicus) with the tape around the body in a horizontal position, with the measurer standing at the side of the participant. Hip circumference (cm) was measured at the widest portion of the buttocks or greatest hip girth (greater trochanter). Body fat distribution was assessed indirectly by the waist-to-hip ratio (WHR). Blood pressure was measured twice, using a standard aneroid sphygmomanometer on the left arm after at least 10 minutes of rest with the average of the measurements recorded.

Specimen collection and storage

The procedure was carried out during the wet season (March–October) at the Metabolic Research Ward, University College Hospital Ibadan. About 12 ml of blood were drawn from each participant (after 10–12 hours of overnight fast) from the antecubital vein, with 4 ml dispensed into an ethelene diamine tetra acetic acid (EDTA) bottle for the fasting lipid profile, 4 ml into a gel clot/activator bottle for other serum lipid studies, 25(OH) vitamin D, hs-CRP, and fasting insulin measurement, and 4 ml into a fluoride oxalate bottle for fasting glucose measurement. Specimen bottles were transported immediately after blood collection in ice packs to the laboratory. Each specimen bottle was centrifuged at 3,000 *g* for 15 minutes within 30 minutes of blood collection. Full clot retraction of the blood sample in the gel clot/ activator bottle was allowed before centrifuging.

Both the serum and plasma were decanted into their respective labeled plain bottles and stored at –20° C for not more than 3 months before assay.

Assay methods

Plasma glucose was measured using the glucose oxidase method on an automated chemistry analyzer (LW C 100 plus). Standard enzymatic methods were also used to analyze plasma TC (cholesterol oxidase), HDL-C (direct HDL PEGME), and TG (lipase/glycerol phosphate-oxidase) on an automated chemistry analyzer (LW C 100 plus). The Friedewald equation was used to calculate LDL-C. Serum 25(OH) vitamin D, hs-CRP, fasting insulin, and Lp(a) levels were manually assayed using the ELISA method, while serum Apo B and Apo A1 were analyzed using immunoturbidimetric assays on an automated chemistry analyzer (LW C 100 plus). The quality control (QC) samples were run with the participants' samples in each assay batch. The test results were accepted only if the QC results of the analytical run were in the recommended range. Inter assay coefficient of variation was determined and was 2.57% for TC, 3.31% for TG, 1.74% for HDL-cholesterol, 3.24% for Apo B, 2.4% for ApoA-1, 2.98% for glucose, 6.37% for insulin, 9.2% for Lp(a), 8.5% for hsCRP, and 7.9% for 25(OH) vitamin D.

Statistical Analysis

Statistical analysis was performed using SPSS version 20.0. Descriptive characteristics for participants were presented as mean ± standard error. Student's *t*-test was used to compare the differences in normally distributed variables between groups (i.e., anthropometric data and clinical data, fasting glucose, and other lipid studies), while Mann–Whitney U was used for non-Gaussian variables comparison between the two groups (i.e., fasting insulin and hs-CRP). Pearson correlation was carried out to estimate the associations between serum 25(OH) vitamin D concentrations (independent variable), anthropometric data, fasting glucose, and other lipid studies (dependent variables), except for fasting insulin and hsCRP, which were examined by Spearman rank correlation. The level of significance was taken to be $p < 0.05$.

Results

Demographic characteristics of the study population

A total of 150 apparently healthy young adults, with ages ranging from 18 to 25 years, were recruited and grouped into two; 57 of the participants fell into Group A category (i.e., those with serum 25 (OH) vitamin D concentrations ≥ 30 ng/ml) and the remaining 93 of the participants fell into Group B category (i.e., those with serum 25 (OH) vitamin D concentrations < 30 ng/ml).

Anthropometric and Clinical data of Group A (Vit D sufficient) and Group B (Vit D insufficient/deficient)

Group A individuals were significantly taller than group B ($p = 0.008$). There was no significant difference in weight, BMI, hip circumference, waist circumference, and waist-hip ratio, systolic and diastolic blood pressure between the two groups. (Table 1)

Comparison of atherogenic markers between Group A (Vit D sufficient) and Group B (Vit D insufficient/ deficient)

The HDL-C was significantly lower in group B compared to group A. There was no significant difference in the concentrations of FPG, fasting insulin, TC, TG, LDL, LP(a), Apo B, Apo A1, and Apo B/Apo A1 ratio and hsCRP when compared between the groups. (Table 2)

Correlation between vitamin D status and risk variables of participants

There was a weak positive correlation between vitamin D status and HDL-cholesterol; and a weak negative correlation between vitamin D status and hsCRP in young adults with serum 25(OH) vitamin D concentrations ≥30 ng/ml. A weak positive correlation also existed between vitamin D status and WC in young adults with serum 25(OH) vitamin D concentrations <30 ng/ml. (Table 3).

Discussion

The present study aimed to assess the relationship between risk markers of atherogenesis and vitamin D status [serum 25(OH) D concentrations] in healthy young adults, in South-Western, Nigeria. It is important to note that results obtained from this study were within reference intervals for all variables assessed [other than vitamin D status]; though some obvious differences were observed between the two groups compared.

It was found that young adults with vitamin D sufficiency (levels ≥ 30 ng/ml) were significantly taller than those with vitamin D deficiency/ insufficiency (levels < 30 ng/ml; similar to the work of Billoo et al. [27] and Muguntan et al. [28]; who

Table 1. Anthropometric and Clinical data of Group A (Vit D sufficient) and Group B (Vit D insufficient/deficient).

Parameters	Group A	Group B	*p* value
	n = 57	*n* = 93	
Height (m)	1.67 ± 0.01	1.64 ± 0.01	0.008*
Weight (kg)	60 ± 1.18	57 ± 0.92	0.055
BMI (m^2/kg)	21.6 ± 0.37	21.3 ± 0.28	0.518
HC (cm)	95 ± 0.88	94 ± 0.77	0.436
WC (cm)	77 ± 0.85	75 ± 0.82	0.110
WHR	0.82 ± 0.01	0.81 ± 0.05	0.057
SBP (mmHg)	114 ± 1.60	111 ± 1.46	0.120
DBP (mmHg)	73 ± 1.24	72 ± 1.04	0.316

**p* value significant at <0.05). Student's *t*-test was used for comparison of parameters.
BMI = Body mass index, HC = Hip circumference, WC = Waist circumference, WHR = Waist hip ratio, SBP = Systolic blood pressure, DBP = Diastolic blood pressure.

Table 2. Comparison of atherogenic risk markers between Group A (Vit D sufficient) and Group B (Vit D insufficient/deficient).

Variables	Group A	Group B	*p* value
	n = 57	*n* = 93	
FPG (mg/dl)	71 ± 1.13	67 ± 1.35	0.070
FSI (uIU/ml)	2.74 ± 0.67	0.85 ± 0.10	0.077^z
TC (mg/dl)	160 ± 4.11	156 ± 2.65	0.470
HDL (mg/dl)	63 ± 1.51	58 ± 1.15	0.007*
TG (mg/dl)	68 ± 2.38	71 ± 2.16	0.374
LDL (mg/dl)	83 ± 3.84	84 ± 2.47	0.853
LP(a) (mg/dl)	71 ± 2.99	72 ± 1.91	0.881
Apo B (mg/dl)	87 ± 2.79	85 ± 2.40	0.518
Apo A1 (mg/dl)	171 ± 2.51	167 ± 1.81	0.215
Apo B/Apo A1	0.51 ± 1.11	0.51 ± 1.31	0.942
hsCRP (mg/l)	6.00 ± 0.60	6.23 ± 0.45	0.822^z

**p* value significant at <0.05; *z* signifies Mann–Whitney U for non-Gaussian variables. Student's *t*-test was used for comparison of variables, except for fasting insulin and hs-CRP in which Mann–Whitney U test was used.
FPG = Fasting plasma glucose, FSI = Fasting serum insulin, LDL = Low-density lipoprotein, HDL = High-density lipoprotein, TG = Triglyceride, Lp(a) = Lipoprotein (a), ApoB = Apolipoprotein B, ApoA-1 = Apolipoprotein A-1, hsCRP = highly sensitive C-reactive protein.

Table 3. Correlation between vitamin D status versus (vs) risk variables of participants.

Vit D status versus	Group A		Group B	
risk variables	*r*	*p*	*r*	*p*
Height (m)	0.44	0.050	0.13	0.290
Weight (kg)	0.15	0.430	0.02	0.550
BMI (kg/m^2)	0.40	0.150	0.07	0.110
WC (cm)	0.53	0.050	0.16	0.040*
HC (cm)	0.49	0.070	0.21	0.410
WHR	0.20	0.470	0.01	0.350
SBP (mmHg)	0.01	0.300	0.31	0.110
DBP (mmHg)	0.15	0.600	0.21	0.550
FPG (mg/dl)	0.30	0.290	0.17	0.210
FSI (uIU/ml)	0.44	0.620	0.21	0.090
TC (mg/dl)	0.36	0.190	0.27	0.220
HDL (mg/dl)	0.41	0.040*	0.50	0.050
TG (mg/dl)	0.25	0.090	0.15	0.230
LDL (mg/dl)	0.41	0.140	0.23	0.070
LP(a) (mg/dl)	0.03	0.013	0.12	0.320
Apo B (mg/dl)	0.38	0.250	0.29	0.220
Apo A1 (mg/dl)	0.11	0.270	0.01	0.150
Apo B/Apo A1	0.39	0.160	0.26	0.440
hsCRP (mg/l)	-0.37	0.010*	0.23	0.070

**p* value < 0.05. Spearman rank correlation was carried out to estimate the associations between serum 25(OH) vitamin D concentrations and fasting insulin and, hs-CRP; while Pearson correlation was used for all other variables.
BMI = Body mass index, HC = Hip Circumference, WC =Waist Circumference, WHR =Waist Hip Ratio, SBP = Systolic blood pressure, DBP = Diastolic blood pressure, FPG = Fasting plasma glucose, FSI = Fasting serum insulin, LDL = Low-density lipoprotein cholesterol, HDL = High-density lipoprotein cholesterol, TG = Triglyceride, Lp(a) = Lipoprotein (a), ApoB = Apolipoprotein B, ApoA1 = Apolipoprotein A1, hsCRP = high sensitive C-reactive protein.

demonstrated that vitamin D supplementation in the young brought about an appreciable gain of weight and height during follow-up. Vitamin D is important for skeletal development and its deficiency or insufficiency may result in short stature associated with rickets in children or osteomalacia in adults [29]. Though the beneficial effect of vitamin D on bone health has been clearly established in the past, there are limited data available on the effect of vitamin D status on linear growth. In an observational study, in which no supplementation was not used, low serum 25(OH) D was associated with slower linear growth in children [30], and, therefore, the present study supports the importance of sufficient vitamin D for linear bone growth.

In the present study, HDL-cholesterol was significantly lower in participants with insufficient/deficient vitamin D status when compared with those with adequate vitamin D status. This is similar to the findings of Smotkin-Tangorra et al [31], who reported that obese children and adolescents with vitamin D deficiency had reduced levels of

HDL-cholesterol. It was also reported by Sonuga et al [32], in a study involving vitamin D supplementation in preeclamptic pregnant women, that there was a significant increase in HDL-cholesterol levels after supplementation, while the deficient group had significantly low levels of HDL-cholesterol. The present study further showed a positive association between vitamin D status and HDL-cholesterol, as in the reports of Karhapää et al. [33] and Jorde et al. [34], supporting existing suggestions from other countries that adequate vitamin D status may assist in the generation of "good" cholesterol.

hsCRP is an objective and sensitive index of overall systemic inflammatory activity [35]. Vitamin D has anti-inflammatory effects which could result in beneficial effects on a variety of disease states associated with inflammation, including diabetes mellitus and cardiovascular disease [36,37]. In the present study, a significant inverse relationship was found between vitamin D status and hsCRP in the participants with adequate vitamin D status as was found by Robinson et al. [38] and Shifa et al. [39]. Both vitamin D deficiency and inflammation have been linked to cardiovascular diseases through the modulatory effect of vitamin D on the RAAS and its inhibitory effect on vascular smooth muscle hypertrophy [40]. Vitamin D mediates its effects in various tissues through its specific receptors, regulating the pro-inflammatory and systemic inflammatory response in the tissues. Activation of vitamin D receptors (VDR) located in the nucleus of the macrophages helps to inhibit the activation of the pro-inflammatory transcription factor; nuclear factor kappa B (NF-κB), which is required for TNF-α expression. This suggests that VDR activation has an intrinsic inhibitory role in inflammation [41,42] and also when there is vitamin D insufficiency, NF-κB activation occurs invariably leading to the endogenous induction of hsCRP. It is also well-known that active vitamin D acts to down-regulate pro-inflammatory cytokines, such as TNF, IL-6, IL-1, and IL-8 [43], which induce CRP production [44]; and that it up-regulates anti-inflammatory cytokine, such as IL-10 concurrently [45], explaining the finding that adequate vitamin D status [25(OH) vitamin D concentrations] was associated with lower concentrations of hsCRP in the present study, likely protecting against the development and progression of CVD.

Conclusion

This present study supports existing evidence from earlier published works that reduced vitamin D status could increase both inflammation and dyslipidemia, which are both well-known risk factors for incident cardiovascular diseases. It also supports earlier data suggesting that adequate vitamin D status could reduce the later risks of overt adult cardiovascular disease.

Recommendation

Vitamin D status [serum 25(OH) D] should be screened in the young periodically and supplements should be given if low concentrations of serum vitamin D are found; and outdoor activities with safe sun exposure should also be encouraged if population repletion cannot be improved by public health measures.

Study Limitations

The sample size for this study is small. Pro/anti-inflammatory markers were not measured, though highly-sensitive C reactive protein, a known marker of overall inflammatory activity in the human body, was measured. Other indicators of inflammation, such as erythrocyte sedimentation rate and white cell count were not assessed.

Authors' contributions

OOS developed the manuscript and performed the laboratory analyses, OOE participated in laboratory analysis, and AAS performed the statistical analysis. All authors read and agreed on the final manuscript.

Competing interests

None declared by authors.

References

[1] Lim SS, Vos T, Flaxman AD, Danaei G, Shibuya K, Adair-Rohani H, et al. A comparative risk assessment of burden of disease and injury attributable to 67 risk factors and risk factor clusters in 21 regions, 1990–2010: a systematic analysis for the Global Burden of Disease Study 2010. Lancet 2012; 380:2224–60.

[2] Seedat YK. Recommendations, for hypertension in sub-Saharan Africa. Cardiovasc J S Afr 2004; 15:157–8.

[3] Kearney PM, Whelton M, Reynolds K, Muntner P, Whelton PK, He J. Global burden of hypertension:

analysis of worldwide data. Lancet 2005; 365:217–23.
[4] Lozano R, Naghavi M, Foreman K, Lim S, Shibuya K, Aboyans, Vet al. Global and regional mortality from 235 causes of death for 20 age groups in 1990 and 2010: a systematic analysis for the Global Burden of Disease Study 2010. Lancet 2012; 380:2095–128.
[5] World Health Organization. Global action plan for the prevention and control of non-communicable diseases: 2013–2020. WHO, Geneva, Switzerland, 2013.
[6] Hojskov CS, Heickendorff L, Moller HJ. High throughput liquid-liquid extraction and LC-MS-MS assay for determination of circulating 25(OH) vitamin D3 and D2 in the routine clinical laboratory. Clin Chem Acta 2010; 411:114–6.
[7] Ruiz-Núñez B, Pruimboom L, Dijck-Brouwer DAJ, Muskiet FAJ. Lifestyle and nutritional imbalances associated with Western diseases: causes and consequences of chronic Vitamin D and cardiometabolic risk. J Nutr Biochem 2012; 24:1183–201.
[8] Ridker P.M., Hennekens C.H. C-reactive protein and other markers of inflammation in the prediction of cardiovascular disease in women. N Engl J Med 2000; 342:836–43.
[9] Polkowska A, Glowinska-Olszewska B, Tobiaszewska M, Bossowski A. Risk factors for cardiovascular disease in children with type I diabetes in 2000–2010 in Podlaise Province. Pediatr Endocrinol Diabetes Metab 2015; 20(2):47–54.
[10] Mbewu AD, Durrington PN. Lipoprotein (a): structure, properties and possible involvement in thrombogenesis and atherogenesis. Atherosclerosis 1990; 85:1–14.
[11] Nordestgaard BG, Chapman MJ, Ray K, Borén J, Andreotti F, Watts GF, et al. A. Lipoprotein (a) as a cardiovascular risk factor: current status. Eur Heart J 2010; 31:2844–53.
[12] Willeit P, Kiechl S, Kronenberg F, Witztum JL, Santer P, Mayr M, et al. Discrimination and net reclassification of cardiovascular risk with lipoprotein(a): prospective 15-year outcomes in the Bruneck Study. Jam Coll Cardiol 2014; 64:851–60.
[13] Sniderman AD, Furberg CD, Keech A, Roeters van Lennep JE, Frohlich J, Jungner I, et al. Apolipoproteins versus lipids as indices of coronary risk and as targets for statin treatment. Lancet 2003; 361:777–80.
[14] Holme I, Aastveit AH, Hammar N, Jungner I, Walldius G. Inflammatory markers, lipoprotein components and risk of major cardiovascular events in 65,005 men and women in the Apolipoprotein MOrtality RISk study (AMORIS). Atherosclerosis 2010; 213:299–305.
[15] Haverkate F, Thompson SG, Pyke SDM, Gallimore JR, Pepys MB. Production of C-reactive protein and risk of coronary events in stable and unstable angina. Lancet 1997; 349:462–6.
[16] Haffner SM. The metabolic syndrome: inflammation, diabetes mellitus, and cardiovascular disease. AM J Cardiol 2006; 97:3A–11A.
[17] Pradhan AD, Manson JE, Rifai N, Buring JE, Ridker PM. C-reactive protein, Interlukin-6, and Risk Developing Type 2 Diabetes mellitus. JAMA 2001; 286:327–34.
[18] Schottker B, Haug U, Schomburg L, Kohrle J, Perna L, Muller H, et al. Strong association of hydroxyvitamin D concentrations with all-cause, cardiovascular, cancer and respiratory disease mortality in a large cohort study. Am J Clin Nutr 2013; 97:782–93.
[19] Christakos S, Dhawan P, Verstuyf A, Verlinden L, Carmeliet G. Vitamin D: metabolism, molecular effects of action, and pleiotropic effects. Physiol Rev 2016; 96:365–408.
[20] Lavie C.J., Lee J.H., Milani R.V. Vitamin D and cardiovascular disease. Will, it, live, up to its hype? J Am Coll Cardiol 2011; 58:1547–56.
[21] Reis AF, Hauache OM, Velho G. Vitamin D endocrine system and the genetic susceptibility to diabetes, obesity and vascular disease: a review of evidence. Diabetes Metab 2005; 31(4):318–25.
[22] Wang TJ, Pencina MJ, Booth SL, Jacques PF, Ingelsson E, Lanier K, et al. Vitamin D deficiency and risk of cardiovascular disease. Circulation 2008; 117:503–11.
[23] Giovannucci E. Expanding roles of vitamin D. JCEM 2009; 94(2):418–20.
[24] Holick MF. Vitamin D: a millennium perspective. J Cell Biochem 2003; 88:296–307.
[25] El-Hajj FG Nabulsi GM, Nabulsi M, Choucair M, Salamoun M, Hajj Shahine C, Kizirian A, et al. Hypovitaminosis D in healthy school children. Pediatrics 2001; 107(4):E53.
[26] Holick MF. Vitamin D status: measurement, interpretation and clinical application. Ann Epidemiol 2009; 19(2):73–8.
[27] Billoo AG, Murtaza G, Memon MA, Khaskheli SA, Iqbal K, Rao MH. Comparison of oral versus injectable vitamin-D for the treatment of nutritional vitamin-D deficiency rickets. J Coll Physicians Surg Pak 2009; 19:428–31.
[28] Mugunthan SR, Rao YK, Midha T, Bajpai A. Effect of vitamin D supplementation on growth parameters of children with vitamin D deficiency: a community based randomized controlled trial. Int J Contemp Pediatr 2017; 4:2070–4.
[29] Pettifor JM. Vitamin D deficiency and nutritional rickets in children. In: Feldman DPW, Glorieux FH (eds.). Vitamin D. 2nd edition, Elsevier Academic Press, Boston, MA, pp 1065–84, 2005.
[30] Hannan MT, Litman HJ, Araujo AB, McLennan CE, McLean RR, McKinlay JB, et al. Serum 25-hydroxyvitamin D and bone mineral density in a racially and ethnically diverse group of men. J Clin Endocrinol Metab 2008; 93:40–6.

[31] Smotkin-Tangorra M, Purushothaman R, Gupta A, Nejati G, Anhalt H, Ten S. Prevalence of vitamin D insufficiency in obese children and adolescents. J Pediatr Endocrinol Metab 2007; 20(7):817–23.
[32] Sonuga A.A., Asaolu MF, Sonuga, OO. Effects of vitamin D supplementation on lipid profile and plasma glucose of preeclamptic women in Ibadan, Nigeria. Open Access Library J 2018; 5:e4410.
[33] Karhapää P, Pihlajamäki J, Pörsti I, Kastarinen M, Mustonen J, Niemelä O, et al. Diverse associations of 25-hydroxyvitamin D and 1, 25-dihydroxy-vitamin D with dyslipidaemias. J Intern Med 2010; 268(6):604–10.
[34] Jorde R, Figenschau Y, Hutchinson M, Emaus N, Grimnes G: high serum 25-hydroxyvitamin D concentrations are associated with a favorable serum lipid profile. Eur J Clin Nutr 2011; 64(12):1457–64.
[35] Kluft C. Identifying patients at risk of coronary vascular disease: the potential role of inflammatory markers. EHJSup 2004; 6:21–7.
[36] Zhang Y, Leung DYM, Richers BN, Liu Y, Remigio LK, Riches DW, et al. Vitamin D inhibits monocyte/macrophage proinflammatory cytokine production by targeting MAPK Phosphatase-1. J Immunol 2012; 188(5):2127–35; doi:10.4049/jimmunol.1102412
[37] Calton EK, Keane KN, Newsholme P, Soares MJ. The impact of vitamin D levels on inflammatory status: a systematic review of immune cell studies. PLoS One 2015; 10(11):e0141770; doi:10.1371/journal.pone.0141770.
[38] Robinson AB, Tangpricha V, Yow E, Gurion R, McComsey GA, Schanberg LE, et al. Vitamin D deficiency is common and associated with increased C-reactive protein in children and young adults with lupus: an Atherosclerosis Prevention in Pediatric Lupus Erythematosus substudy. Lupus Sci Med 2014; 1:e000011.
[39] Shifa K, Jithesh TK, Mirshad P, Supriya-Simon A. Vitamin D deficiency and inflammatory marker in subjects with dyslipidaemia. Am J Biochem 2017; 7(1):6–9.
[40] Levin A, Li YC. Vitamin D and its analogues: do they protect against cardiovascular disease in patients with chronic kidney disease? Kidney Int 2005; 68(5):1973–81.
[41] Chen MH, Colan SD, Diller L. Cardiovascular disease: cause of morbidity and mortality in adult survivors of childhood cancers. Circ Res 2011; 108(5):619–28.
[42] Mishra A, Liu S, Sams GH, Curphey DP, Santhanam R, Rush LJ, et al. Aberrant over expression of IL-15 initiates large granular lymphocyte leukemia through chromosomal instability and DNA hypermethylation. Cancer Cell 2012; 22:645–55.
[43] Sharma A, Satyam A, Sharma JB. Leptin, IL-10 and inflammatory markers (TNF-α, IL-6 and IL-8) in pre-eclamptic, normotensive pregnant and healthy non-pregnant women. Am J Reprod Immunol 2007; 58:21–30.
[44] Hvilsom GB, Thorsen P, Jeune B, Bakketeig LS. C-reactive protein: a serological marker for preterm delivery? Acta Obstet Gynecol Scand 2002; 81:424–42.
[45] Lau SY, Guild SJ, Barrett CJ, Chen Q, McCowan L, Jordan V, et al. Tumor necrosis factor-alpha, interleukin-6, and interleukin-10 levels are altered in preeclampsia: a systematic review and meta-analysis. Am J Reprod Immunol 2013; 70:412–27.